Appendix A: Assessment Methodology demonstration. (EPA ESA Maine WQS consultation BE)

Although the methodology uses the dwarf wedgemussel as an example of the application to ammonia and cadmium effects, the methodology is applied similarly for the Atlantic salmon, using appropriate data.

1. Effect Assessment Methodologies

1.1 Acute Effect Assessment Methodology: Residential Exposure Effects

The protectiveness of the freshwater acute ammonia and freshwater acute cadmium criteria magnitudes was assessed by identifying or estimating acute toxicity values (i.e., LC_{50}, which is the concentration that will kill half of the test population) for Maine aquatic listed species that were then adjusted to represent protective low effect threshold concentrations as described below. Acute toxicity values used to develop the acute effects assessments were obtained from Appendix A of their respective 304(a) aquatic life criteria documents (Ammonia, USEPA 2013; Cadmium, USEPA 2016) and were specifically used to derive the acute criterion (i.e., bold values in Appendix A of USEPA 2013 and underlined values in USEPA 2016). These data were identified from EPA's ECOTOX database, the open and grey literature, and have been subjected to extensive data quality review (see USEPA 1985 for data quality). Acute ammonia values have been normalized to a pH of 7 (all freshwater animals) and 20°C (freshwater invertebrates only), consistent with criteria derivation (USEPA 2013). Acute cadmium toxicity data have been normalized to a total hardness of 100 mg/L as $CaCO_3$ consistent with criteria derivation (USEPA 2016). Ideally, species-specific toxicity data for listed species of concern are available to support an acute effects assessment; however, data limitations (i.e., no available data for a specific species) often required use of surrogate toxicity data.

EPA considered acute toxicity data at the closest taxonomic level available to determine geometric mean acute toxicity values for each species assessed. Considering surrogate toxicity data at the most phylogenetically-related taxonomic level possible accounts for genetically-derived traits conserved across taxa that may directly influence species and taxa sensitivity to a pollutant. Species-specific and surrogate acute toxicity data obtained from Appendix A of USEPA (2013) and USEPA (2016) represent sensitivity expressed as a concentration that will kill half of the test population (i.e., LC_{50}). EPA then transformed the acute toxicity data (expressed as LC_{50}) to an acute low effect threshold concentration that is not expected to kill more than five percent of a listed species population (i.e., LC_5) under continuous exposure conditions. Representing acute low effect thresholds as LC_5 values is conservative considering high-quality toxicity tests are considered acceptable when demonstrating up to 10 percent lethal effects in control (unexposed to pollutant) organisms, typically resulting from natural mortality. That is, at effect levels below ten percent, it is often difficult to distinguish whether or not observed responses are the effect of natural morality or pollutant exposures themselves.

Raw acute toxicity data may be used to calculate LC_5 values directly from the concentration-response (C-R) curves of the listed species-specific toxicity tests, when available. However, not all acute tests provide concentration-response data. Therefore, species-specific, or surrogate LC_{50} values (which represent listed species 50% effect level), were transformed to an acute low effect threshold concentration through the use of an acute taxonomic adjustment factor (TAF) or an acute mean adjustment factor (MAF). An acute TAF was calculated by averaging (geometric mean) the ratios of LC_{50}:LC_5 from ammonia or cadmium toxicity tests conducted using species in the closest possible phylogenetic proximity (same species, genus, family, or order) as the listed species that is being assessed. When data availability did not allow for the development of an acute TAF within the same order as the species being assessed, EPA applied an acute invertebrate or vertebrate TAF (depending on whether the listed species assessed was an invertebrate or vertebrate). The acute invertebrate TAF and the acute vertebrate TAF were calculated as the geometric mean of genus-level LC_{50}:LC_5 ratios of invertebrates and vertebrates, respectively. An acute MAF was used to adjust species effect concentrations (i.e., LC_{50}) to low effect threshold concentrations (i.e., LC_5), when 1) an acute TAF is not available within the same order as the listed species being assessed; and 2) when the acute invertebrate TAF and the acute vertebrate TAF were not significantly different via a two-sample t-test assuming unequal variances ($\alpha = 0.05$). The acute MAF is calculated as the geometric mean of all genus-level LC_{50}:LC_5 ratios available. Acute invertebrate and vertebrate TAFs and the acute MAF are calculated as the geometric mean of their respective genus-level LC_{50}:LC_5 ratios to limit the influence of LC_{50}:LC_5 ratios from species that are overly represented in a dataset, similar to criteria derivation (USEPA 1985).

Listed species-specific or surrogate LC_{50} values were then divided by an appropriate adjustment factor (i.e., acute TAF or acute MAF depending on data availability) to derive an acute low effect threshold concentration. Dividing LC_{50} values by an adjustment factor to identify a low-level effect concentration is an approach that is fundamentally similar to acute criteria derivation[1], but is more specific to the chemical and species assessed. Acute low effect threshold concentrations were then compared to corresponding criteria magnitudes (i.e., criterion maximum concentration [CMC]) to assess potential adverse effects of ammonia or cadmium exposures at the acute criterion concentration over conservative exposure durations.

Vertebrate sensitivity to ammonia is dependent on pH, with tolerance decreasing as pH increases. Invertebrate sensitivity is influenced by both pH and temperature, with invertebrate tolerance increasing as temperature decreases (see USEPA 2013). When salmonids in the Genus *Oncorhynchus* are present, EPA's recommended acute criterion magnitude is limited to protect adult rainbow trout (a commercially and recreationally important species), which are the most sensitive species at lower temperatures (i.e., < 15.7°C). Thus, the CMC is both pH- and temperature-dependent. The CMC increases with decreasing temperature as a result of increased invertebrate insensitivity until it reaches a plateau of 24.10 mg TAN/L at 15.7°C and below, where the most sensitive taxon is the temperature invariant rainbow trout. Unlike the criterion magnitude, however, invertebrate sensitivity to ammonia continues to decrease as temperature decreases. Figure 1-1 depicts the change in the ammonia CMC across water chemistries (i.e., the

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[1]The Final Acute Value (FAV; fifth centile of genus mean acute values) is divided by 2.0 to derive the Criterion Maximum Concentration (CMC). The FAV was divided by 2.0 to ensure the CMC is representative of a concentration that will result in low level effects (e.g., 0-10%) to the 5th percentile of sensitive genera. EPA's 1978 proposed guidelines for deriving criteria (43 Fed. Reg. 21425, 21506-21518 (May 18, 1978)) outlined the derivation of a generic LC_{50} to LC_{low} (i.e., 0-10% effect) adjustment factor of 0.44 (or divide by 2.27). The adjustment factor of 2.27 was derived as the geometric mean of the quotients of the highest concentration that killed 0-10% of the organisms divided by the LC_{50} in 219 acute toxicity tests. The geometric mean adjustment factor (2.27) outlined in the 1978 proposal was subsequently rounded to 2.0 in EPA's final 1985 Guidelines (USEPA 1985).

change in acute criterion magnitude with temperature at pH 6, 7, 8, and 9), and how the acute low effect threshold for dwarf wedgemussel changes with the criterion magnitude proportionally (factor difference of 2.664) until 15.7°C. The acute effects assessment was developed using toxicity data normalized to reference conditions (pH = 7, temperature = 20°C) and compared to the corresponding CMC in those same reference conditions. Because species sensitivity and the CMC both change similarly across water chemistries, conclusions based on reference conditions translate to other water chemistries.

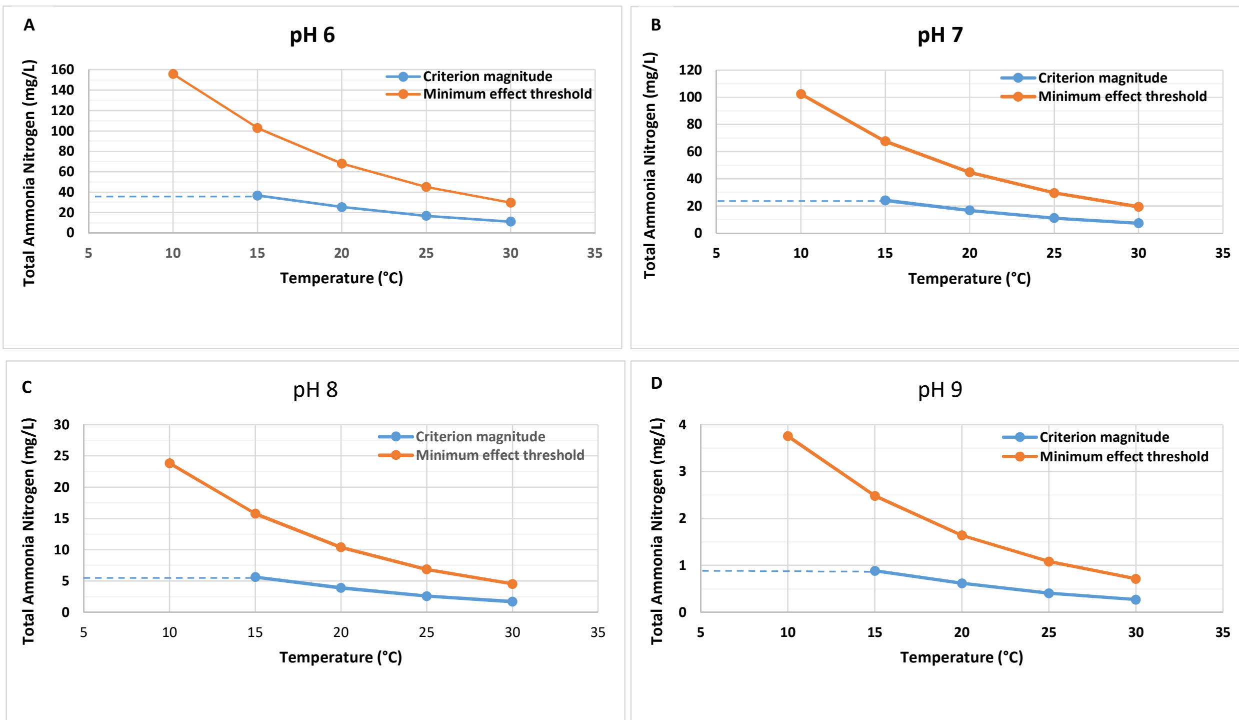

Figure 1-1. Acute ammonia criterion magnitudes extrapolated across a temperature gradient at pH 6, 7, 8 and 9 (Panels A-D)[2].

In contrast to ammonia, species sensitivity to cadmium is only dependent on water hardness, with tolerance increasing as hardness increases (see USEPA 2016). The CMC increases with increasing hardness across the range of hardness in typical ambient surface water (acute toxicity hardness slope 0.9789). Figure 1-2 depicts the change in the cadmium CMC across water hardness of 25 to 400 mg/L as $CaCO_3$, and how the acute low effect threshold for dwarf wedgemussel changes with the criterion magnitude proportionally (factor difference of 14.94). The acute effects assessment was developed using toxicity data normalized to a reference condition (hardness = 100 mg/L) and compared to the corresponding CMC in those same

[2] The acute low threshold concentration calculated for dwarf wedgemussel (per Section 2.2.1 of this document) is overlaid on the acute criterion magnitude. Using the depiction at pH 7 (Panel B) as the example, the criterion magnitude increases with decreasing temperature as a result of increased invertebrate tolerance until the CMC reaches a plateau of 24.10 mg TAN/L at 15.7°C and below, where the most sensitive taxa is the temperature invariant rainbow trout. The criterion plateau (indicated by the dotted line) changes with pH. The dwarf wedgemussel acute low effect threshold continues to decrease as temperature decreases. The factor difference between the acute criterion magnitude and acute low effect threshold for dwarf wedgemussel is 2.664.

reference conditions. Because species sensitivity and the CMC both change similarly across water chemistries, conclusions based on reference conditions translate to other water chemistries.

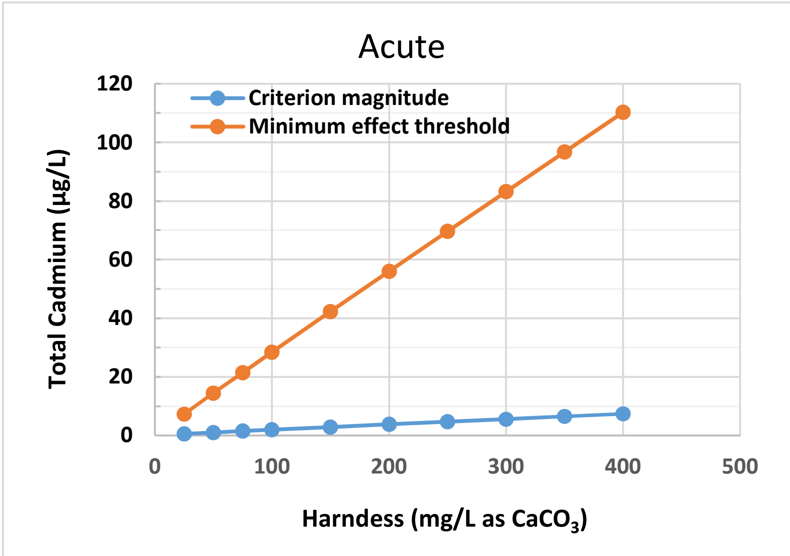

Figure 1-2. Acute cadmium criterion magnitudes extrapolated across a gradient of water hardness, overlaid with the dwarf wedgemussel acute low effect threshold concentration[3].

Assessing a criterion magnitude alone does not consider the duration and frequency components of the criterion and represents an overly conservative exposure scenario that assumes a pollutant concentration in all Maine freshwaters will be at the acute criterion magnitude indefinitely. If a listed species' acute low effect threshold concentration is greater than the corresponding acute criterion magnitude, then a refined assessment and consideration of the criterion duration and realistic exposure is not necessary, and approval of the acute criterion is not likely to adversely affect that particular listed species through acute effects in freshwaters.

1.2 Chronic Effect Assessment Methodology: Residential Exposure Effects

The protectiveness of the chronic freshwater ammonia and chronic freshwater cadmium criteria magnitudes was assessed by identifying or estimating chronic toxicity values for Maine aquatic listed species that were then adjusted to represent protective low effect threshold concentrations as described below. Ammonia chronic toxicity values used to develop the chronic effects assessments were obtained from Appendix B of the ammonia 304(a) aquatic life criteria document (USEPA 2013) and cadmium chronic toxicity data were obtained from Appendix C of the cadmium criteria document (USEPA 2016). These data were identified from EPA's ECOTOX database, the open and grey literature, and have been subjected to extensive data quality review (see USEPA 1985). Chronic ammonia toxicity data (i.e., EC_{20}) used to support the effects assessment have been normalized to a pH of 7 (all freshwater species) and 20°C (freshwater invertebrates only), consistent with criteria derivation (USEPA 2013); and chronic cadmium toxicity data have been normalized to a total hardness of 100 mg/L as $CaCO_3$, consistent with criteria derivation (USEPA 2016).

[3] The cadmium criterion magnitude and the dwarf wedgemussel acute low effect threshold both increase with increasing water hardness. The factor difference between the acute criterion magnitude and acute minimum effect threshold for dwarf wedgemussel is 14.94.

Ideally, species-specific toxicity data are available to support a chronic effects assessment; however, similar to acute toxicity discussed above, data limitations often required use of surrogate toxicity data. Unlike acute criteria derivation, which typically uses a generic LC_{50} to LC_{low} adjustment factor (i.e., 2.0[1] -- see footnote 2, above; USEPA 1985), the chronic criteria are based directly on chronic effect concentrations (i.e., EC_{20} which represents a 20 percent effect/inhibition concentration) [USEPA 1985]. Because a concentration that results in chronic effects to 20% of an exposed listed species population could be considered excessively high, EPA developed adjustment factors to transform EC_{20} to EC_5 to represent a chronic low effect threshold concentration following an approach similar to the acute effects assessment methodology discussed above.

Raw chronic toxicity data may be used to calculate EC_5 values directly from the concentration-response (C-R) curves of the listed species-specific toxicity tests, when available. However, not all chronic tests provide concentration-response data. Therefore, species-specific, or surrogate EC_{20} values (which represent listed species 20% effect level), were transformed to a chronic low effect threshold concentration through the use of a chronic taxonomic adjustment factor (TAF) or a chronic mean adjustment factor (MAF). A chronic TAF was calculated by averaging (geometric mean) the ratios of EC_{20}:EC_5 from ammonia and cadmium toxicity tests, respectively, conducted using species in the closest possible phylogenetic proximity (same species, genus, family, or order) as the listed species that is being assessed. When data availability did not allow for the development of a chronic TAF within the same order as the species being assessed, EPA applied a chronic invertebrate or vertebrate TAF (depending on whether the species assessed was an invertebrate or vertebrate). The chronic invertebrate TAF and the chronic vertebrate TAF were calculated as the geometric mean of genus-level EC_{20}:EC_5 ratios of invertebrates and vertebrates, respectively. A chronic MAF was used to adjust species effect concentrations (i.e., EC_{20}) to low effect threshold concentrations (i.e., EC_5), when 1) a chronic TAF is not available within the same order as the listed species being assessed; and 2) when the chronic invertebrate TAF and the chronic vertebrate TAF were not significantly different via a two-sample t-test assuming unequal variances ($\alpha = 0.05$). The chronic MAF is calculated as the geometric mean of all genus-level EC_{20}:EC_5 ratios available. Chronic invertebrate and vertebrate TAFs and the chronic MAF are calculated as the geometric mean of their respective genus-level EC_{20}:EC_5 ratios to limit the influence of EC_{20}:EC_5 ratios from species that are overly represented in a dataset, similar to criteria derivation (USEPA 1985).

Listed species-specific or surrogate EC_{20} values were then divided by an appropriate adjustment factor (i.e., chronic TAF or chronic MAF depending on data availability) to derive a chronic low effect threshold concentration. Chronic low effect threshold concentrations were then compared to the corresponding criterion magnitude (i.e., criterion continuous concentration [CCC]) to assess potential adverse effects of ammonia or cadmium exposures at the chronic criterion concentration.

The ammonia CCC is both pH- and temperature-dependent. Chronic vertebrate sensitivity to ammonia is affected by pH, while chronic invertebrate sensitivity to ammonia is influenced by temperature and pH. Figure 1-3 depicts the change in CCC across water chemistries and how the chronic low effect threshold for dwarf wedgemussel changes proportionally with the criterion magnitude (factor difference of 1.024). Because species sensitivity and the CCC both change similarly across water chemistries, conclusions based on reference conditions translate to other water chemistries.

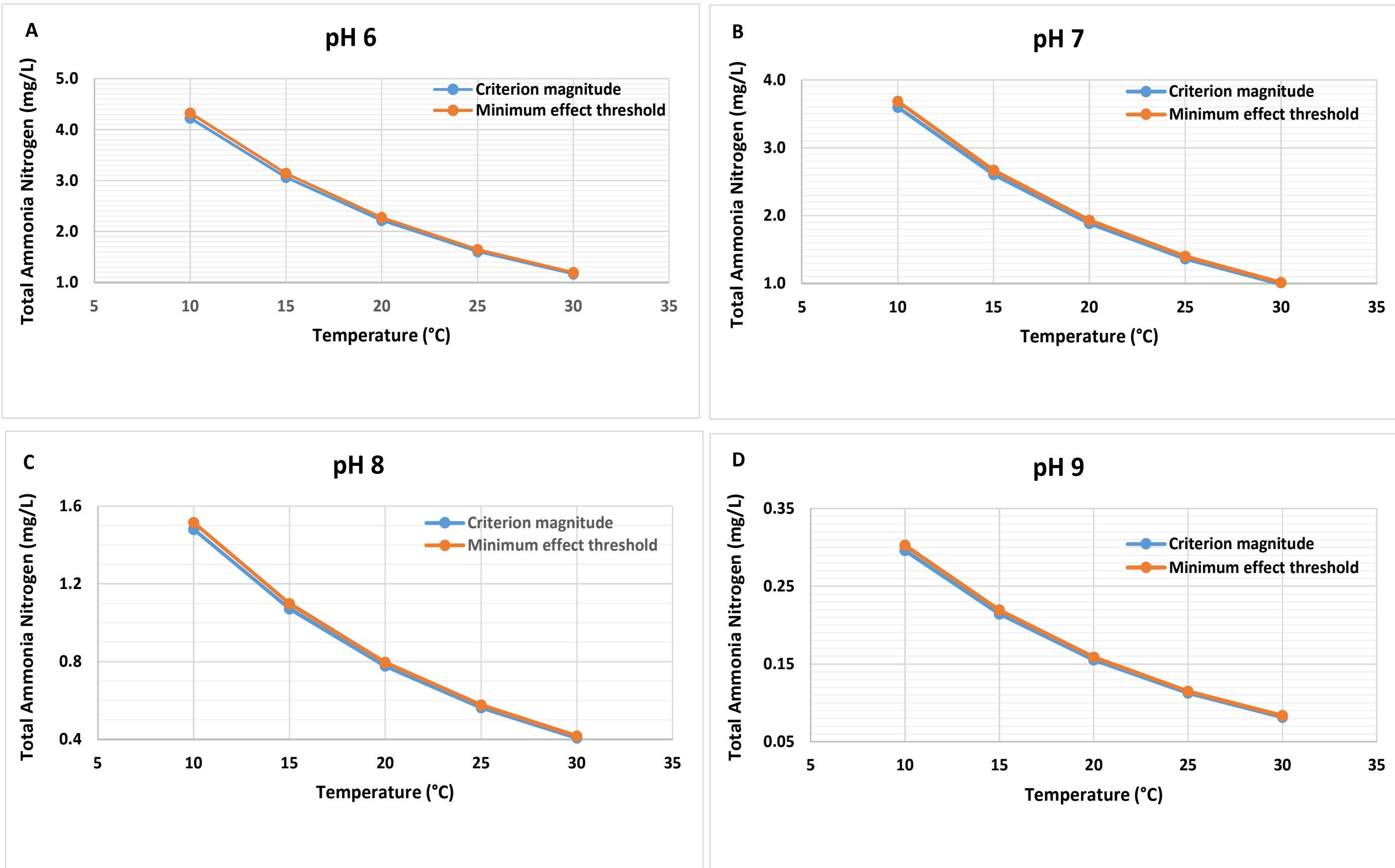

Figure 1-3. Chronic ammonia criterion magnitude extrapolated across a temperature gradient at pH 6, 7, 8 and 9 (Panels A-D) with the dwarf wedgemussel chronic ammonia low effect threshold concentration overlaid[4].

In contrast to ammonia, species sensitivity to cadmium is only dependent on water hardness, with tolerance increasing as hardness increases (see USEPA 2016). The CCC increases with increasing hardness across the range of hardness typical of natural ambient surface water, but with a slightly lower slope than for the CMC (chronic toxicity hardness slope 0.7977). Figure 1-4 depicts the change in the cadmium CCC across water hardness of 25 to 400 mg/L as $CaCO_3$, and how the chronic low effect threshold for dwarf wedgemussel changes with the criterion magnitude proportionally (factor difference of 9.5). The chronic effects assessment was developed using toxicity data normalized to a reference condition (hardness = 100 mg/L) and compared to the corresponding CCC in those same reference conditions. Because species sensitivity and the CCC both change similarly across water chemistries, conclusions based on reference conditions translate to other water chemistries.

[4] The factor difference between the chronic criterion magnitude and chronic low effect threshold for dwarf wedgemussel is 1.024.

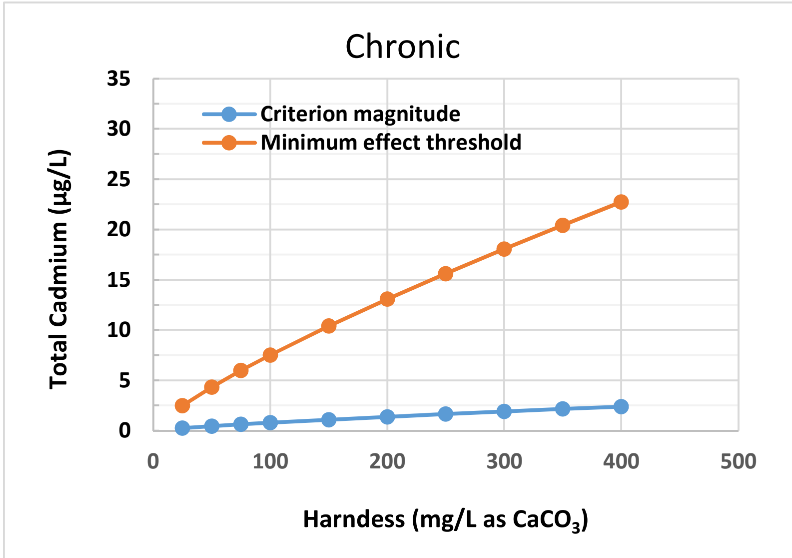

Figure 1-4. Chronic cadmium criterion magnitudes extrapolated across a gradient of water hardness overlaid with the dwarf wedgemussel chronic low effect threshold concentration[5].

Assessing a criterion magnitude alone does not consider the duration and frequency components of the criterion and represents an overly conservative exposure scenario that assumes a pollutant concentration in all Maine freshwaters will be at the chronic criterion magnitude indefinitely. If a listed species' chronic low effect threshold concentration is greater than the corresponding chronic criterion magnitude, then a refined assessment and consideration of the criterion duration and realistic exposure is not necessary, and approval of the chronic criterion is not likely to adversely affect that particular listed species through chronic effects of residential exposure in freshwaters.

1.3 Other Effects (All listed species): Assessment of Acute and Chronic Criteria

Following assessment of acute and chronic effects of residential exposure on listed aquatic species, EPA considered and assessed potential other (non-residential) effects of the water quality standard approval actions on the aquatic and aquatic-dependent listed species. To assess those potential effects, EPA considered conservatisms associated with criteria derivation and implementation as well as potential effects to listed species prey items, and on ingesting drinking water.

[5] The criterion magnitude increases and the dwarf wedgemussel chronic low effect threshold both increase with increasing water hardness. The factor difference between the chronic criterion magnitude and chronic low effect threshold for dwarf wedgemussel is 9.5.